IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of SHAW et al.	Confirmation No: 8619
Application No. 10/810,388	Examiner: GUCKER, Stephen
Filed: March 26, 2004	Group: 1649

For: ASSESSING NEURONAL DAMAGE FROM BLOOD SAMPLES

DECLARATION UNDER 37 C.F.R. §1.132

Commissioner for Patents
P. O. Box 1450
Alexandria, VA 22313-1450

I, Dr. Gerry Shaw, declare as follows:

1. I am a named inventor in patent application 10/810,388 entitled "ASSESSING NEURONAL DAMAGE FROM BLOOD SAMPLES." I am currently a full time employee of the University of Florida College of Medicine. I am a Professor of Neuroscience, Anatomy and Cell Biology in the McKnight Brain Institute.

2. I hold a doctorate in Zoology from the University of London, King's College, as described in more detail in my curriculum vitae (c.v.) appended hereto. Since 1975 I have worked regularly in the field of cellular neurobiology, with much of my work focused on neurofilament proteins. I have authored and co-authored 98 peer reviewed publications, including the only single author monograph on neurofilaments, a topic of obvious relevance to this application.

3. I have reviewed the Office Action dated May 30, 2008 and the prior art

references cited therein.

4. I have reviewed the following independent claim which, as amended in the Response Under 37 C.F.R. 1.111 filed herewith, is recited in the present invention. This claim is copied below and appears in italics:

Claim 1: A method of detecting neuronal injury in a subject, the method comprising the steps of:

(a) providing a blood, serum, or plasma sample from the subject;

(b) contacting the blood, serum, or plasma sample with an antibody that specifically binds to NF-H in the sample;

(c) detecting the presence or amount of NF-H in the sample,

wherein NF-H can be detected in quantities as low as 50 pg; and

(d) correlating the presence or amount of NF-H in the sample with the neuronal injury.

5. Hu et al. was determined by the examiner to render the claimed invention unpatentable when combined with Zemlan in addition to Grainger et al. or Posmantur et al. According to the examiner, "[i]t would have been obvious to one of ordinary skill in the art at the time of the invention to use the methods of Hu with the blood samples of Zelman because it is simpler and easier to procure a blood sample and assay it by ELISA than it is to procure a CSF sample by lumbar puncture because the blood sample can be simply taken from the arm (no usual side effects) while the CSF sample needs to be taken from the spinal cord region with the attendant risks of damaging the cord and then producing the side effects usually resulting from lumbar puncture such as headaches," "the use of chicken polyclonal antibodies in combination with the previously applied art

would have yielded predictable results to one of ordinary skill in the art at the time of the invention" and "it would be obvious to try to look for the missing NF-H protein from the brain in the blood within a few hours of a neuronal injury as taught by Postmantur with the methods disclosed by Hu and Zelman, and the combination with the previously applied art would have yielded predictable results to one of ordinary skill in the art at the time of the invention." I respectfully disagree with these determinations for the reasons explained below.

6. Developing the claimed invention was not straightforward or obvious over the prior art. My coinventor Dr. Brian Pike and I demonstrated for the first time that NF-H can be robustly detected in blood, plasma, and serum of injured mammals using the claimed invention. We disagree with the examiner's assertions that this was an obvious step based on prior art. Even if one could argue that in principle it is obvious that a brain-specific protein might be detectable in blood following CNS injury, it would not be obvious to succeed. We now know that most human genes are expressed in the brain, so in principle almost any gene product could be a viable biomarker. The trick is knowing which of the numerous possible candidates to focus on, then having an assay of sufficient sensitivity and specificity to detect this protein, and having access to appropriate experimental and control blood samples from animals or patients on which to perform and validate this assay. Many brain-specific proteins are not resistant to proteases, and following injury, are either not present in blood, serum, or plasma at all or are present at levels that cannot be detected using heretofore available methods. In contrast to the heretofore available methods, the claimed invention provides for detecting a brain-specific protein that is resistant to proteases from a blood sample with heretofore

unreported sensitivity. Simply extrapolating from findings made in CSF by Hu et al. and others would not result in the claimed invention, as many CNS-derived proteins detectable in CSF have not been reported to be also detectable in blood. CSF is within the brain, adjacent to the source of damaged neural cells and potential brain injury biomarkers and typically contains only about 50μg/ml total protein, more than three orders of magnitude below the average level of protein in blood, which is about 60mg/ml. In addition, the total volume of CSF in an adult human is ~150mls, about one thirtieth the total volume of blood, typically about 5 liters. A brain-specific protein in CSF is therefore expected to be reduced in relative concentration by several thousand-fold (i.e. 1/30 X 50/60,000) assuming it moves from the CSF to the blood without being degraded, modified or sequestered en route. Such a protein could go directly from a serious injury site to the blood, but even in this case the large volume and very high protein content of blood result in rapid and very significant dilution of the protein. Blood also contains proteases and other enzymes which may degrade, modify and/or sequester the protein. These facts therefore make CSF a vastly more favorable environment in which to detect brain injury biomarkers (as taught by the prior art). Combining the teachings of Hu et al. with the teachings of Zemlan, Grainger et al. and Posmantur et al. would not result in the sensitive assay that we developed (and that is currently claimed) for detecting neuronal injury in a subject by detecting NF-H in the subject's blood, serum or plasma at quantities as low as 50 pg.

In fact, the assay used by Hu et al. is very poorly described and characterized. The capture antibody used was a rabbit antibody called R61d which was raised against a mixture of NF-L, NF-M and NF-H. Presumably R61d binds all three proteins, though no

further Western blotting or other information is given in Hu et al. or other publications from this group. Assuming it does bind all three proteins, it is clearly not likely to capture NF-H as efficiently as an assay based on antibody to NF-H alone, such as the one we developed. Significantly, since the assay was never calibrated with pure NF-H, the actual amounts of NF-H being detected in control and affected CSF are not known, the Hu et al. data being presented only as "relative levels". Binding in the assay was not shown to be inhibited by preincubation with pure NF-H and the assay has never been shown to work on blood samples. Finally Hu et al. amplified the signal by using an unusual bienzyme substrate-recycle ELISA, suggesting that the NF-H signal was not robust even on CSF samples. Even if one of skill in the art thought to apply the assay of Hu et al. to blood samples, it does not appear likely that it would have been workable with this particular assay.

Regarding Zemlan, this reference includes no data regarding neurofilament proteins – from any bodily fluid. Zemlan describes experiments involving only tau protein. Tau and NF-H are entirely different proteins, belonging to two distinct and ancient protein families with quite different domain organizations, functions, and binding properties. Zemlan provides no working examples involving NF-H, and when combined with Hu et al., Posmantur et al., or Grainger et al., would not result in the claimed invention. There were actually three serious potential problems which could have made our invention unworkable. Firstly, it was possible that NF-H either does not enter the blood, enters the blood but rapidly degrades, or enters the blood and becomes inaccessible as it binds to one or other blood proteins or is rapidly removed by filtration in the kidneys or other means. Secondly, even on entry to blood in an accessible form,

NF-H might still not have been detectable given the low amount present and the high background of blood proteins. The development of an assay with high enough avidity, specificity and sensitivity to allow detection of this protein in blood was therefore an essential feature of the discovery. Thirdly, it was important to have access to a collection of well described control and experimental blood samples from animals in which the NF-H blood signal could be strongly and unambiguously detected.

Given these serious potential problems, we therefore had no likely reason to expect that we would be able to detect NF-H in a blood, serum or plasma sample as a result of neuronal injury, as recited in the instant claims. We regarded the pilot experiments we performed as having a low likelihood of success, but, if successful, having a very high potential impact. The experiments were only feasible since we were able to overcome the three difficulties listed above and also because neurofilaments have been a major focus of our research over many years, and we have access to both the necessary purified proteins and many different NF-H antibodies. We were quite surprised when our preliminary experiments showed that NF-H could in fact be robustly detected in blood samples from animals with experimental CNS injuries. Unlike the highly soluble and monomeric tau protein described in Zemlan, neurofilaments are the most stable components of the axon, and are found in a very high molecular weight 10nm diameter filament protein complex which can be readily isolated by simply centrifuging them out of a cell homogenate made in the presence of detergents. This procedure works since neurofilaments form a well defined and very high molecular weight 3 dimensional structure even in the absence of a plasma membrane (e.g. Shaw and Hou. J. Neurosci. Res. 25:561-568 1990). Most of the other components of the axon, including tau, are

soluble under these conditions. This neurofilament protein complex is held together by alpha-helical coiled coil interactions, known to be very stable under physiological conditions, particularly in molecules of this kind (Mason and Arndt. ChemBioChem. 5:170-176 2004). Neurofilaments and their subunits are thought, based on several lines of evidence, to be the longest lived and most stable proteins in the neuronal cytoplasm, and were known to be quite resistant to cellular proteases. It was therefore not obvious that any of the neurofilament subunits or possibly fragments derived from them would be released from damaged axons in measurable amounts. The work of Posmantur et al, cited by the examiner, shows that the amount of NF-H *remaining* in damaged regions of the CNS declines following injury, and that less NF-H is lost in the presence of calpain protease inhibitors. In fact, addition of the calpain inhibitor reduced the loss of NF-H from ~65% to a much more modest ~20% (Posmantur et al. figure 2). It is important to note that Posmantur et al. used a Western blotting assay to measure the amount of the *intact* 200kDa NF-H band. The loss of the band only shows that the NF-H protein has been proteolytically cleaved, and does not necessarily indicate that it is no longer in the brain tissue assayed and so may have migrated to the CSF and/or blood. Posmantur et al. do not show data on lower molecular weight NF-H fragments which could have addressed this issue. Figure 1 of the same paper shows that calpain inhibition almost completely prevents the loss of the NF-L protein band in the same paradigm. Since NF-L and NF-H are part of the same protein complex, the neurofilament, this finding suggests that *neither* protein is actually being lost from the brain in this experiment in the presence of calpain inhibitors. There is also no reason to think that in the absence of calpain inhibitors the neurofilament subunits are being released from damaged neurons, this issue

is simply not addressed in Posmantur et al. In summary, the Posmantur et al. reference says nothing whatsoever about how much, if any, of the injury induced reduction in NF-H is *not* due to cytoplasmic degradation. In addition, some loss of brain NF-H in this experiment could be due to localized extracellular degradation in the CNS, reduced synthesis of NF-H which is known to occur after injury, release of NF-H into the CSF or, finally, release into blood. Since there are clearly many other ways to account for the loss of NF-H following injury, there seems to be no strong *a priori* reason to think that a significant fraction would find its way to the blood. In fact our recent work shows that contusion injuries to the rat brain similar to those made by Posmantur et al. generally release only very small amounts of NF-H into the blood over the 24 hour time period studied by this group, and that the signals detected significantly overlap with those obtained from control animals (see figure 1, Anderson et al. J. Neurotrauma 25:1079-1085 2008). More robust levels of blood NF-H were seen in blood 48 hours after injury, beyond the time scale of the Posmantur et al. experiments. I feel, therefore, that for several reasons, Posmantur et al., when combined with Hu et al. and Zemlan, simply does not teach or suggest that a search for NF-H in blood would be a useful experiment. In addition, even with the availability of a good NF-H ELISA, an experiment performed over the same time course as Posmantur et al. would have given equivocal results.

Several features of NF-H are at odds with those of other known biomarker molecules, including tau. The NF-H subunit is, at 200kDa SDS-PAGE molecular weight, much larger than the ~30kDa tau protein. Surely the fact that NF-H is much larger and much less soluble than tau makes it much more surprising that this molecule would find its way into blood, plasma and serum. We believed that it was of course possible that NF-

H would be detectable in blood following CNS injury, or we would not have looked for it. However, for the several reasons outlined above, it did not seem likely or "obvious" that it would be present in detectable amounts.

7. There is no evidence in the prior art, published papers or patents, that neuronal injury can be assessed by detecting NF-H in a blood, plasma or serum sample in quantities as low as 50 pg as claimed in the present application, or in fact at any level. The conventional wisdom at the time the application was filed was that proteins capable of leaving the brain following brain injury or degeneration would have to be low molecular weight and soluble proteins in order to cross the blood-brain barrier. As noted above, NF-H is neither low molecular weight nor soluble. Those of skill in the art at the time the application was filed thought that future biomarkers were likely to be similar to the most widely used CNS injury and degeneration biomarker, the calcium binding protein S100β which is expressed heavily in astrocytes. This protein has a molecular weight of only 11kDa. Other potential brain injury biomarker proteins, all of which have somewhat questionable utility, are much smaller than NF-H (e.g. Myelin basic protein 18kDa, tau ~30kDa, neuron specific enolase ~50kDa, glial fibrillary acidic protein ~50kDa). Based on the prior art, the much larger NF-H molecule does not look like a promising candidate, and appears to be, to date, the highest molecular weight blood biomarker known. Our published immunoblotting data shows that NF-H in CSF of patients recovering from brain aneurysms is largely intact and undigested (Lewis et al. J. Cerebral Blood Flow and Metabol. 28:1261-1271 2008), and as yet unpublished data show that the NF-H signal in blood has a gel filtration molecular weight of ~600kDa,

suggesting that the molecule is multimeric or at least part of a very high molecular weight complex. Clearly these findings, when viewed in light of the prior art and conventional wisdom, are rather surprising and seem to argue against the likely and "obvious" use of NF-H as a blood, plasma and serum biomarker of CNS injury and degeneration.

8. I am the owner and chief executive officer of EnCor Biotechnology Inc. and since March 2007 we have been selling ELISA kits for detecting neuronal damage as claimed in the application. This kit is the only one on the market apart from the version sold by Millipore, which uses key antibody reagents obtained from EnCor (see below). We have sold over 100 kits and have generated ~$40,000 in sales revenue without any advertising except through our internet site and published work. A consortium of European labs working on Multiple Sclerosis, led by Gavin Giovannoni (Bart's and London NHS, UK) has expressed their desire to order 40 NF-H ELISA kits, and several smaller orders are being processed. We are confident that sales will increase as our newer publications illustrate further applications for this technology.

9. My biotechnology company, EnCor Biotechnology, exclusively licensed the technology claimed in the present application from the University of Florida, with the right to sublicense to other companies. The technology was licensed to Millipore for a down payment of $20,000 and 32.5% royalties on sales. EnCor supplies NF-H antibody coated ELISA plates and other reagents and Millipore packages them into kits for resale. BioVendor, a company in the Czech republic which sells many ELISAs, bought $20,000 worth of antibodies and protein standards in order to validate the assay and modify it to use in their standard kit format. They have now agreed to terms of a $20,000 down

payment and $100 per kit sold and have ordered enough antibody reagents to make 100 ELISA kits. Another company, Banyan Biomarkers, recently indicated a firm intention to license the technology for use in human diagnosis of traumatic brain injury, and negotiations are scheduled for early December 2008. About two years ago Magellan Biosciences made an offer to buy EnCor Biotechnology outright, partly to gain access to this technology. In every case EnCor was approached by these other companies based on scientific publications, presentations at research congresses, or articles in the biotechnology industry press (see below) and did not solicit any of these business interactions. It seems likely that these business interactions will increase, particularly as more publications describing new applications of the technology appear, and describing new NF-H assays based on novel monoclonal antibodies perhaps more amenable to mass production methods and the generation of more advanced assays.

10. To date, eight federal and private foundation grants have been awarded to myself and various collaborators to further use the technology claimed in the application. The Amyotrophic Lateral Sclerosis (ALS) Association has awarded two of these grants, the VA Medical Center has awarded two, the National Institutes of Health (NIH) has awarded two, the Bethlehem Griffiths Research Foundation awarded one, and I personally was awarded a competitive travel grant, the Alan and Maria Myers Traveling Fellowship. The ALS Association funded this work since we were able to show that NF-H is detectable in the blood of transgenic mice and rats with an ALS like disease state before they become symptomatic of the disease, and the blood NF-H levels increase as the disease state progresses, while the protein was not present in the blood of control mice. Using blood NF-H levels might therefore be usable in drug discovery in these

rodents. The levels of blood NF-H also increase in ALS patients, allowing easy monitoring of axonal degeneration. This will provide new details of the ALS disease process, aid in classifying patients and determining likely prognosis, and allow an assessment of the effectiveness of future drug therapies. The VA proposals look at blood NF-H levels in animal models and patients with various kinds of brain and spinal cord injuries. The NIH proposals funded to date look at blood levels of NF-H in patients with optic neuritis, Leber's hereditary optic neuropathy and other forms of optic nerve disease. The Bethlehem Griffiths Research Foundation is an Australian funding agency which is interested in the application of our work to patients with Multiple Sclerosis. Finally, the traveling fellowship paid for my airplane ticket, living and lab expenses and allowed me to work in the prestigious Howard Florey Institute of the University of Melbourne, Australia. I stayed there for two months, conducting experiments directly associated with the technology claimed in the present application. This fellowship paid $30,000 and has resulted in one published peer reviewed publication so far (Gresle et al. J. Neurosci. Res. 86:3548-3555 2008), and more are expected as the collaboration is ongoing and local funding has been obtained.

Several additional grant proposals centered on this technology are pending, including three to the NIH, a further proposal to the ALS Association, one to the American Health Assistance Foundation and one to the National Multiple Sclerosis Society. Other proposals are at earlier stages of preparation, including further studies aimed at human traumatic brain injury, stroke, Multiple Sclerosis, Parkinson's and Alzheimer's disease. These fourteen successful and pending proposals produced to date ALL make use of the basic NF-H assay and/or second generation assays based on our

original discovery. The work described in and claimed in the present patent application has clearly opened numerous avenues for further research in animals and humans, and much of the work points at future clinical utility. NF-H appears to have much better utility as an axonal injury biomarker than tau protein, and this is clear to both collaborators and funding agencies. In fact, five of our current six publications describe NF-H in blood, serum and plasma, and all report positive correlations in the levels of NF-H with aspects of the damage or disease process studied.

Obtaining extramural funding has become quite a challenge in recent years, so the repeated success of proposals based on the subject matter of the application is a testament to the interest in and potential scientific and clinical impact of this technology.

11. Because the claimed invention is promising and of potential clinical utility, several descriptions of it have made their way to the various media. For examples, see Reuter's Health Report – Professional Medical News 11-04-2005, the November 2005 issue of the online journal Diagnostics Intelligence (only available online to subscribers) , the January 2006 issue of Physician's Weekly, downloadable from; (http://www.physiciansweekly.com/article.asp?issueid=314&articleid=2950) and the March 31 2007 issue of Biotech Transfer Week, which can be downloaded from; (http://www.biotechtransferweek.com/issues/1_5/features/139266-1.html). There have also been several newspaper articles and items on local and national television. For one still accessible example see the "Health Alert" on WISTV of South Carolina; the actual item is no longer on line although all the text of this item can still be downloaded from; http://www.wistv.com/Global/story.asp?S=4624476&nav=0RaS.

In addition, I have encountered such widespread interest in this technology that I am working with more than 20 collaborators in the US and abroad to measure the levels of blood and in some cases CSF NF-H in a variety of CNS damage and disease states. These collaborators include Dr. Axel Petzold MD PhD (Institute of Neurology, London, CSF and blood samples from a variety of CNS damage and disease states), Dr. Gavin Giovannoni MD PhD (Bart's and the London NHS, UK, interested in tracking Multiple Sclerosis progression), Dr. Rick Odland MD (blood from patients with Bell's Palsy), Dr. David Borchelt PhD (University of Florida, blood from ALS and other model mice), Dr. Johnathan Glass MD (Emory University, CSF and blood from patients with familial forms of ALS), Drs. Kevin Boylan MD and Elizabeth Schuster MD (Mayo Clinic Jacksonville, CSF and blood from patients with sporadic ALS and MS), Dr. Neill Graff-Radford MD (Mayo Clinic Jacksonville, blood from Alzheimer's patients), Dr. Mike Weiss MD (University of Florida, blood from pediatric stroke patients), Dr. John Guy MD (Bascom Palmer Eye Institute, Miami, blood from patients with various kinds of optic neuropathy), Dr. Kevin Anderson PhD (University of Florida, blood rat models of traumatic brain injury and spinal cord injury), Dr. Cristina Morganti-Kossman PhD (Alfred Hospital, Melbourne, blood from traumatic brain injury patients), Dr. Bevyn Jarrott PhD (Howard Florey Institute, Melbourne, blood from animal models of stroke), Drs. Trevor Kilpatrick MD PhD, Melissa Gresle PhD and Helmut Buetzheuven MD PhD (Howard Florey Institute, Melbourne, blood from animal models of and patients with MS), Dr. James Vickers PhD (University of Hobart, Tasmania, blood from Alzheimer's and MS patients), Dr. Hiroaki Kamishina DVM (Iwate University, Japan, CSF and blood from dogs with spinal cord injuries and demyelinating diseases), Dr. Roger Clemmons

DVM (University of Florida, CSF and blood from dogs with spinal cord injuries and demyelinating diseases). Dr. Clair Ringger DVM and Dr. Steeve Giguere (both in private practice, but associated with the University of Florida Vet School, blood from new born foals with hypoxic ischemic encephalopathy), Dr. Brian Blyth MD and Dr. Jeff Bazarian MD (University of Rochester, N.Y., blood from traumatic brain injury patients), Dr. Dena Howland PhD (VA Medical Center, blood from animal models of spinal cord injury), Dr. Paul Hoffman MD (VA Medical Center, blood from MS patients), Dr. Stephen Lewis MD, (University of Florida, CSF and blood from patients recovering from aneurysmal subarachnoid hemorrhage and traumatic brain injury), Dr. Felice A Wener MD (University of Tennessee, blood biomarkers of pediatric traumatic brain injury) and Dr. Juan Solano MD (University of Miami, CSF and blood from pig models of traumatic brain injury). The fact that more than half of the collaborators are MDs reflects the potential clinical implications of this work. We are discussing several other collaborations which will likely significantly swell this list in the next year. This will include formal collaborations with further groups focusing on human traumatic brain and spinal cord injury, Parkinson's disease, Alzheimer's disease and hemorrhagic and ischemic stroke. In addition, several private biotechnology companies have asked EnCor to run NF-H assays on a fee for service basis.

The research work performed to date has resulted in six peer-reviewed publications, with two more submitted for publication and several others in preparation. (Shaw et al. BBRC 336:1268-1277 (2005); Petzold, and Shaw. J. Immunol. Mets, 319:34-40 (2007); Lewis et al. J. Cerebral Blood Flow and Metabol. 28:1261-1271 (2008); Gresle et al. J. Neurosci. Res. 86:3548-3555 (2008); Anderson et al. J.

Neurotrauma 25:1079-1085 (2008); Guy et al. Mol. Vision (in press, 2008). Like obtaining grant funding, publication requires peer review and is only possible with novel data of some interest and significance. The journals we have published in are among the higher impact in their areas, with the Journal of Cerebral Blood Flow and Metabolism being one of the world's best in this area of research. This paper studied patients with aneurysmal subarachnoid hemorrhage and has generated considerable interest as the profiles of NF-H release into blood and CSF proved to be predictive of patient outcome and also clearly showed that several aspects of the recovery process could be monitored using our blood test. The work also revealed novel features of the recovery process. As one anonymous reviewer stated "The authors findings... are of interest and might be even seminal". The Gresle et al. paper shows that blood NF-H levels accurately reflect the seriousness of disease in experimental allergic encephalomyelitis (EAE) mice, which are a widely used model of human Multiple Sclerosis. We also showed that a drug which ameliorates the EAE disease state also greatly reduced the blood NF-H levels. This finding shows that the monitoring of blood NF-H levels can be used for drug discovery in animal models of human neurodegenerative disease. This may prove to be a very significant finding which may directly impact patients with a variety of such disorders. The Guy et al. paper shows that NF-H is detectable in the blood of patients with Leber's Hereditary Optic Neuropathy and the levels detected are informative of patient outcome. Finally, I have been asked to write a review of the nervous system injury and degeneration biomarker field for the International Journal of Clinical and Experimental Pathology. In conclusion, our unexpected results contrast with the relatively modest findings made with the only other published biomarker of axonal injury, tau, (described

in Zemlan) and strongly suggest that NF-H determination will prove to be much more versatile and informative.

12. In summary, developing the claimed invention was not straightforward or obvious over the prior art. In addition to commercial success, I have encountered significant interest from biotechnology companies, funding agencies, the media, and many collaborators worldwide. I believe that this level of interest reflects the uniqueness and apparent utility of the invention.

13. I further state that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with my knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under §1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

11/25/08

Date

Gerry Shaw, Ph.D.

BIOGRAPHICAL SKETCH (November 2008)

Name	Gerard P. J. Shaw.
Date of Birth	24 March 1953, Nottingham, England.
Citizenship	British/U.K. citizen. Currently permanent resident (green card holder) in the USA.
Marital Status	Married to Regina Hasse, (born 5 October 1957, Duisberg citizen of Federal Republic of Germany) in Göttingen, Federal Republic of Germany, on 18 October 1985.
Family	Daughter Elizabeth, born 26 April 1988, Gainesville, Florida. Son Michael, born 21 January 1992, Gainesville, Florida.
Education	B.Sc. in Zoology, University College London, Department of Zoology, July 1975. Ph.D. in Zoology, King's College London, Department of Biophysics, April 1980.
Academic Address	Department of Neuroscience McKnight Brain Institute University of Florida College of Medicine, Box J-100422, Gainesville, Florida 32610. Tel (352) 294 0037 (lab) Tel (352) 294 0038 (office) Tel (352) 871 8153 (mobile) Fax (352) 392 8347 (lab) Email shaw@ufbi.ufl.edu http://www.mbi.ufl.edu/~shaw http://www.mbi.ufl.edu/Dept/Faculty/Shaw.html
Commercial Address	EnCor Biotechnology Inc. Suite 40, 4949 SW 41st Boulevard Gainesville, Florida 32608 Tel: (352) 372 7022 Fax: (352) 372 7066 Email:admin@encorbio.com http://www.encorbio.com/

Professional Experience

10/75-2/80	Graduate Student, Department of Biophysics, King's College London, England.
3/80-12/81	Postdoctoral Fellow, Max Planck Institute for Biophysical Chemistry, Göttingen, Federal Republic of Germany.
1/82-2/86	Staff Member, Max Planck Institute for Biophysical Chemistry, Göttingen, Federal Republic of Germany.
3/86-8/91	Assistant Professor, University of Florida College of Medicine, Gainesville, Florida, USA.
8/91-8/96	Associate Professor with tenure, University of Florida College of Medicine, Gainesville, Florida, USA.
8/96-present	Full Professor with tenure, adjunct Professor of Anatomy and Cell Biology, University of Florida, College of Medicine, Gainesville, Florida, USA.

12/02-present	Founder and owner of 88% of stock of EnCor Biotechnology Inc., a start-up biotechnology company located from 12/02 to 12/06 in Alachua, Florida, from 12/06 in Gainesville, Florida.

Societies, past and present

Member of Society for Neuroscience.
Member American Society for Cell Biology.
Member Federation of American Societies for Experimental Biology.
Member British Society for Cell Biology.
Member of the American Physiological Society.

Manuscript Reviewing

A total of more than 250 papers including papers submitted to the following journals;
American Journal of Pathology
Annals of Neurology
Alcoholism: Clinical and Experimental Research
Anatomy and Embryology
Biological Bulletin (Woods Hole)
Biotechniques
Biochemistry
Brain Research
Brain Research Bulletin
Cell Motility and the Cytoskeleton
Comments on Developmental Biology
Developmental Neuroscience
Experimental Neurology
Experimental Cell Research
FEBS letters
Gene
Investigative Ophthalmology and Visual Science
Journal of Alzheimer's Disease
Journal of Biological Chemistry
Journal of Cell Biology.
Journal of Cell Science
Journal of Histochemistry and Cytochemistry.
Journal of Neurobiology
Journal of Neurochemistry
Journal of Neurological Sciences
Journal of Neurology, Neurosurgery and Psychiatry
Journal of Neuroscience
Journal of Neuroscience Research
Journal of Neurology, Neurosurgery & Psychiatry
Molecular Biology of the Cell
Molecular Vision
Neuroscience Letters
Neurochemistry International
Protein and Peptide Letters
Regulatory Peptides
Trends in Biochemical Sciences

Book and Book Chapter Reviews

Reviewer for Elsevier of "Gene Transfer and Expression in Mammalian Cells". Edited by S. Makrides
Reviewer for Oxford University Press of of Oxford University Press book "Cell Signalling" by J. T. Hancock (2nd Edition).

Editorial Boards

Currently on editorial board of the International Journal of Clinical and Experimental Pathology.

Service

Grant reviewer for Alzheimer's Association on 5 occasions.
Grant reviewer for Amyotrophic Lateral Sclerosis (ALS) Association.
Grant reviewer for NIH (AG-1 NLS-1, NLS-2, NS) on 7 occasions.
Grant reviewer for NSF (Cell Biology, Neuroscience, Developmental Neuroscience, Neuronal and Glial Mechanisms, and Molecular Biochemistry Programs) on 7 occasions.
Grant reviewer of the Wellcome Trust, London, England, on 4 occasions.
Grant reviewer for MRC of Great Britain.
Grant reviewer for MRC of Canada.
Grant reviewer for Malcom Randall VA Medical Center, Gainesville.
Grant reviewer for NEUC subcommittee, VA Medical Center, Washington DC.

Administration

Member, Search committee for Assistant Professor in Molecular biology, University of Florida Department of Anatomy and Cell Biology 1986-1987.
Member, University Biotechnology Committee, 1988-90.
Chairman, Search committee for Assistant Professor in Molecular Neurobiology, University of Florida Department of Neuroscience 1987-1988.
Member, Interdisciplinary Training Program in Cell and Tissue Biology committee,1989-1992.
Member, M.D./Ph.D. graduate student program committee 1993-1999.
Member, Search committee for Chairman of Anatomy and Cell Biology, University of Florida, 1994-1995.
Member, Core curriculum committee for interdepartmental program in basic medical sciences, College of Medicine, University of Florida, 1995-2001.
Director, Advanced Graduate Program in Neurosciences, University of Florida Health Sciences 1995-2001.
Director, IDP Core curriculum, Metabolism/Signal transduction section, 1996-1999.
Director, IDP Core curriculum, Cytoskeleton/Cell Adhesion section, 1996-1997.
Director, IDP Core curriculum, Signal Transduction/Cancer section, 2000-2001.
Director, IDP Core curriculum, Signal Transduction 2002-present.
Member, Center for Structural Biology faculty advisory committee, 1996-present.
Distinguished judge, UF Medical Guild Graduate Student Research 1996.
Member, Advisory Committee to Associate Dean for Graduate Education, 1996-present.
Judge, UF College of Pharmacy, Research showcase and awards recognition day.
Distinguished judge, UF Medical Guild Graduate Student Research 1997.
Distinguished judge UF Medical Guild Graduate Student Research 1998.
Graduate Coordinator for Neuroscience Department 1998-2001.
Member IDP curriculum committee 1999-2000.
Webmaster for Neuroscience Department and Graduate Program, 1999-2003.
Also general administrator, scientific officer and webmaster for my start up company, 2003-present.

Teaching Experience

1986-1992; "Molecular Neurobiology", (GMS 7731), lecturer 1986, in addition course director 1987-1991. This was an annual graduate level course with a strong emphasis on molecular aspects of the nervous system. I introduced the course with lectures on basic techniques of Biochemistry, Immunology and Molecular Biology. I then lectured on the mechanisms of neurite outgrowth, axon/dendrite differentiation, axonal transport, growth factors in the nervous system, the neuronal cytoskeleton, molecular neuropathology, cancer and oncogenesis, neural adhesion molecules, protein phosphorylation in the nervous system and the structure and function of membrane channels and neurotransmitter receptors. The course also covered the regulation of gene expression, extracellular matrix, signal transduction and second messengers, learning and memory, neuroimmunology and neurogenetics. I usually gave at least 14 hours of lectures, attended most of the other lectures (~30) and organized student presentations and exams for this course.

1993-1995; "Cellular and Molecular Neurobiology", (GMS 7733), course director and lecturer. This graduate level course was the result of fusing the above course with our graduate level Neurophysiology course, covering the same material as previously but now including lectures on membrane physiology, impulse conduction, myelination, excitatory amino acids toxicity and synaptic plasticity. I gave a total of 14 Hours of lectures for this course, ran four 2 hour journal club presentations and discussions and ran one 2 hour lab demonstration.

1987-1995; "Developmental and Systems Neurobiology", (GMS 7736) lecturer. I lectured on control of cell division in early development, the development of antero-posterior and dorso-ventral polarity in Drosophila, and on homeotic genes in Drosophila and mammals.

1986-1995; "Cell and Tissue Research", (GMS 5621), lecturer. I lectured on various aspects of the cytoskeleton including microfilaments, intermediate filaments and microfilament and microtubule-associated motor proteins.

1986-1990 and 1995; "Medical Neuroscience", (GMS 7706C), lecturer and laboratory teacher. Annual 5 week intensive course for medical, dental and graduate students. I have lectured on several topics in this course, presented summaries of parts of the course and taught essentially full time in the laboratory for the 5 weeks that this course runs. The laboratory teaching consists of CNS anatomy, histology and functional connectivity. I have also updated several chapters of our teaching text for this course.

1991-1994; "Dental Neuroscience", lecturer and laboratory teacher. Annual 8 week not quite so intensive course covering the same material as the medical course described above, but in slightly less detail. I lectured in this course and taught CNS anatomy, histology and functional connectivity in the laboratory.

1995-present; As part of the University of Florida's change over to a interdisciplinary program (IDP) in Basic Biomedical Sciences, I became a committee member for production of Basic Sciences Core Curriculum. I also headed the Neuroscience component of the graduate program from 1995 to 2001.

1996-present; Organized and lectured in two sections of GMS 6001 and 6002, the Interdisciplinary Program in Biomedical Science core course for first year graduate students. For the first two years I organized and lectured in two modules of this core course, namely "Metabolism and Signal Transduction" and "Cytoskeleton and Extracellular Matrix". Following some changes to the IDP in 1999 I became co-organizer and lecturer in a new core course section "Signal Transduction and Cancer". I also gave lectures and ran discussions in other modules of this core course. Following more reorganization I became responsible for a section "Signal Transduction", from 2002 to the present.

1997-present; Organizer or co-organizer and lecturer in GMS 6051 "Signal Transduction", 1 Unit IDP advanced course offered as part of Neuroscience and Physiology/Pharmacology IDP advanced concentrations.

1997-present; Organizer or Co-organizer and lecturer in GMS 6074 "Comparative Neurobiology", 2 Unit IDP advanced course in Neuroscience IDP advanced concentrations.

1997-present; Organizer and lecturer in GMS6079 "Computers in Biology", 1 Unit IDP advanced course in Neuroscience IDP advanced concentrations. Since 2005 I have given all 15 lectures in this course. This course has become very popular and since 2007 has been run every year.

1996-2001; Organizer of GMS6029 "Brain Journal Club", I Unit IDP advanced course in Neuroscience IDP advanced concentrations.

2002-present, "Molecular Pharmacology" (GMS 6563), 1 Unit IDP advanced course in the Physiology/Pharmacology, lecturer on Fluorescence Resonance Energy Transfer, course organizer Dr. David Silverman.

Graduate Students

Jeffrey M. Harris, Graduate student (M.D./Ph.D program), Ph. D awarded July 1992. Thesis "Structural and immunological analysis of the carboxyterminal tails of the high molecular weight neurofilament subunit proteins NF-M and NF-H". Ph. D. awarded August 1992. Jeff published a total of 3 scientific papers with me (2 first author) and also one completely on his own. He was a resident in Pediatrics,

University of California, San Francisco for several years, but in 2003 became Senior Clinical Diagnostics Scientist at Genentech, San Francisco.

Laura D. Errante, Graduate student, Ph.D awarded June 1994. Thesis "Identification and characterization of neurofilament-associated proteins", Ph.D. awarded May 1994. Laura published 2 first author scientific papers. Laura is now a Post-Doctoral Fellow in the lab or Dr. Paul Forscher, Yale University.

Kulandar Subramanian, Graduate student, Department of Chemical Engineering University of Florida, Co-mentored by Atul Narang and Gerry Shaw, Thesis "", Ph.D. awarded February 2005. Kulandar published 3 papers as a graduate student, and from his graduate studies after he graduated, and is now a post-doctoral fellow in Harvard Medical School.

Silas Morse Graduate student, produced 3 publications, including one as first author, but choose to take a job in the software industry in the summer of 2006.

Ved Prakash Sharma, Graduate student, Department of Chemical Engineering, University of Florida, Co-mentored by Atul Narang and Gerry Shaw, graduated in 2007. Ved published 2 papers as a graduate student, one as first author. Ved started a post doctoral position in New York University in fall 2007.

I am currently a committee member for ~10 other graduate students.

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Commentaries

My work has been discussed numerous times in news articles in Nature, Science and other journals. For examples see Nature 300:580-581 (1982), The Scientist 12:10 (1998) and Molecular Therapy 5: 654 (2002). Paper 22 is listed as a "Milestone Paper" on the Alzheimer's Research Forum website (http://www.alzforum.org/pap/powsearch.asp). Papers 40, 46 and 47 were each evaluated as of "outstanding interest" in Current Opinions in Structural Biology 4:912-918 (1994), Current Opinions in Cell Biology 7:183-189 (1995) and Current Opinions in Neurobiology 4:742-751 (1994) respectively. 4 other papers have been evaluated as "of interest" or "of special interest". The data in paper 88 was the subject of several online articles including Reuter's Health Report, the 11/05 issue of the online journal Diagnostics Intelligence, the January 2006 issue of Physician's Weekly and several spots in newspapers and on television. For a recent example see the "Health Alert" on WISTV of South Carolina, the item being downloadable from http://www.wistv.com/Global/story.asp?S=4624476&nav=0RaS. Also see Biotech Transfer Week, March 31 2007 issue.

Citation Record

A search of the Science Citation Index as of September 19 2008 reveals that research papers and reviews on which I am author have received a total of at least 4911 citations. The most cited papers on which I am first author are numbers 1 (158), 6 (301), 8 (347), 11 (184), 13 (101), 21 (175), 32 (158), 39 (97), 53 (232) and 73 (92). The most cited papers on which I am co-author but not first author are number 2 (140), 7 (182), 22 (375), 45 (91), 46 (414), 47 (86) and 60 (92).

Seminars, Invited Presentations etc.

1) EMBO workshop, "Cell Motility" invited participant, April 1979.
2) Max Planck Institute for Psychiatry, Munich, Federal Republic of Germany, invited by Dr. Gerhard Isenberg, April 1981.
3) EMBO workshop "Intermediate Filaments", Ulm, Federal Republic of Germany, invited speaker, April 1982.
4) EMBO workshop "The Molecular Mechanisms of Nervous System Development", Strassbourg, France, invited speaker, June 1982.
5) Max Planck Institute for Brain Research, Niederrad, Frankfurt, Federal Republic of Germany, invited by Dr. Heinz Wässle, October 1982.

6) Case Western Reserve University, Department of Anatomy, Cleveland, Ohio, invited by Dr. Ray Lasek, February 1983.
7) Max Planck Institute for Biophysical Chemistry, Department of Physiology, invited by Dr. Owen Hamill, June 1983.
8) University of Pittsburg, Department of Neuroscience, Pittsburg, invited by Dr. Ray Lund, June 1983.
9) Washington University, Department of Anatomy and Cell Biology, Saint Louis, invited by Dr. Mark Willard, June 1983.
10) Harvard Medical School, Department of Neurobiology, Boston, invited by Dr. Ursula Dräger, June 1983.
11) Wood's Hole Marine Biological Laboratory, Wood's Hole, New England, invited by Dr. David Potter, June 1983.
12) New York University School of Medicine, Department of Pharmacology, New York, invited by Dr. Ron Liem, June 1983.
13) Columbia University, Department of Neurobiology, New York, invited by Dr. Leland Ellis, June 1983.
14) University of California, San Francisco, Department of Biochemistry, invited by Dr. Regis Kelly, June 1983.
15) Max Planck Institute for Biophysical Chemistry, Department of Neurochemistry, invited by Dr. John Walker, August 1983.
16) Max Planck Institute for Biophysical Chemistry, Department of Molecular Biology, invited by Dr. Tom Jovin, January 1984.
17) University of Bielefeld, Department of Developmental Biology, Bielefeld, Federal Republic of Germany, invited by Dr. Harald Jockusch, February 1984.
18) University of Florida, Department of Neuroscience, Gainesville, invited by Dr. William Luttge, March 1984.
19) Albany Medical College, Department of Anatomy, Albany, New York, invited by Dr. Gary Banker, April 1984.
20) University of Virginia, Department of Surgery, Charlottesville, invited by Dr. Oswald Steward, April 1984.
21) Purdue University, Department of Biological Sciences, West Lafayette, Indiana, invited by Dr. Meredith Appleberry, May 1985.
22) "Intermediate Filaments: Structure, Function and Pathology" Meeting at Irsee, Federal Republic of Germany, invited speaker, 1985.
23) University of Florida, Whitney Marine Biological Laboratory, Marineland, Florida, invited by Dr. Paul Linser, June 1985.
24) Rudolf Magus Institute for Pharmacology, and Institute for Molecular Biology, State University of Utrecht, Holland, invited by Dr. Willem Gispen, December 1985.
25) South Eastern Nerve Net Meeting, invited speaker March 1987.
26) Baylor College of Medicine, Department of Biophysics, Houston, invited by Dr. Kimon Angelides, 1988.
27) International Society for Developmental Biology Meeting, Israel, invited speaker (unable to attend) June 1988.
28) University of Florida, Department of Physiology, invited by Dr. Mohan Raizada 1989.
29) University of Florida, Neuroscience departmental seminar, January 1991.
30) The Miami Project to Cure Paralysis, Miami, invited by Dr. Scott Whittemore, March 1992.
31) Mount Sinai School of Medicine, New York, invited by Dr. James Vickers, June 1992.
32) University of Connecticut, Department of Neuroscience, Storrs, invited by Dr. Enrico Mugnaini, June 1992.
33) The University, Dundee, Department of Anatomy and Cell Biology, invited by Dr. Birgit Lane, June 1993
34) ASCB/EMBO "Intermediate filaments" meeting, Airlie House Virginia, invited speaker June 1993.
35) University of Florida, Department of Pharmacology and Therapeutics, invited by Dr. Jeffrey Harrison, September 1994.
36) ASCB annual meeting San Francisco, "Protein-protein interactions in cell signaling", minisymposium, Invited speaker December 1994.
37) University of Florida, Neuroscience departmental seminar, January 1995.
38) Colorado State University, Neuroscience Program, Fort Collins, invited by Dr. James Bamburg, February 1995.
39) University of Colorado, Department of Biochemistry, Boulder, invited by Dr. Mike Klymkowsky, February 1995.

40) Emory University, Department of Physiology, Atlanta, invited by Dr. Ron Abercrombie, March 1995.
41) South Eastern Nerve Net Meeting, Scientific organizer and session chairman, March 1996.
42) Institute of Psychiatry, De Crespigny Park, London, invited by Dr. Brian Anderton, August 1996.
43) University of Florida, Whitney Marine Biological Laboratory, Marineland, Florida, invited by Steve Munger, September 1996.
44) "Antibodies: Tools in Molecular Sciences", Workshop and symposium, University of Florida, invited by Dr. Paul Klein, May 1997.
45) University of Texas Medical Branch, Galveston, invited by Dr. Owen Hamill, Sept 1999.
46) Mayo Clinic Jacksonville, Florida, invited by Dr. Dennis Dickson, May 2001.
47) University of Florida, Department of Anatomy and Cell Biology, invited by Dr. Chris West, June 2001.
48) Montreal General Hospital, McGill University, Quebec, Canada, invited by Dr. Jean-Pierre Julien, March 2003.
49) University of Florida, McKnight Brain Institute, Neuroscience departmental seminar Oct 2003, invited by Dr. Ron Mandel.
50) University of Florida, McKnight Brain Institute, presentation to McKnight Foundation board, Feb 2004.
51) Institute of Neurology, Queen Square London, invited by Dr. Axel Petzold. July 2005.
52) Gainesville Area Innovation Network (GAIN), Gainesville, invited by Patti Breedlove, July 2005.
53) 4th International Conference on "Biochemical Markers for Brain Damage", September 2005, invited speaker.
54) Annual Society for Neuroscience Meeting, Washington, October 2005, invited speaker.
55) Research presentation to the Department of Chemistry, University of Florida, January 2006, invited by Dr. Weihong Tan.
56) Department of Physiological Sciences, College of Veterinary Medicine, January 2006, invited by Dr. Kevin Anderson.
57) University of Florida, Whitney Marine Biological Laboratory, invited by Dr. Leonid Moroz., May 2006.
58) Newman Hall, University of Melbourne, Australia, invited by Dr. Bevyn Jarrott, August 2006.
59) Howard Florey Institute, University of Melbourne, Australia, invited by Dr. Ernest Jennings, September 2006.
60) Annual Society for Neuroscience Meeting, Atlanta, Georgia, October 2006, invited speaker.
61) ALS initiative meeting/scientific workshop, Cambridge, Massachusetts May 2007, invited by Lucie Bruijn.
62) 7th Cerebral Vascular Biology International Conference and Biomarker satellite meeting, Ottawa, Ontario, invited speaker, June 2007.
63) Mayo Clinic Jacksonville Florida, invited speaker at UF/Mayo retreat, December 2007.
64) University of Florida, McKnight Brain Institute, Neuroscience departmental seminar, invited by Dr. Lucia Notterpek, March 2008.
65) University of Rochester Medical School, Rochester, New York, invited by Dr. Brian Blyth, June 2008.
66) University of Florida, Center for Nano-Bio Sensors, University of Florida, invited by Dr. Maria Palazuelos, July 2008.
67) University of Florida, Parkinson's and movement disorders group, invited by Dr. Hubert Fernandez, November 2008.

Funding History

NIH RO1 NS22695; "Neurofilament modifications: extent and significance"; Principle investigator G. Shaw, (30% time/effort), funding from 3/1/1986-6/30/1997. ~$780,000 direct costs.

NIH RO1 AG07470-01; "Aberrant protein ubiquitinylation in aged and Alzheimer brain"; Principle investigator V. Chau, Co. P. I. G. Shaw (15% time/effort), 12/1/1987-11/30/1990, ~$300,000 direct costs.

Department of Sponsored Research Development Award (University of Florida); "Microinjection of proteins and nucleic acids into cultured neurons". Principle investigator G. Shaw. 7/1/1987-6/30/1988. $15,467.

NIH RO1 NIH RO1 GM44823; "Neuronal MAP interactions with microtubules"; Principle investigator Dr. Dan Purich. Co-P.I. Dr. G. Shaw (10% time effort), funding from 4/1/91 to 3/31/95, ~$1,025,000 direct

costs.

Department of Sponsored Research Development Award (University of Florida); "Ultrastructural analysis of paired helical filaments" P. I. Dr. G. Shaw, $4,800, 5/1/91-4/30/92.

NIH Small Instrumentation Program; "Application of a confocal imaging system"; Principle investigator Dr. G. S. Bennett, Dr. G. Shaw, Co-P.I. Funding for $50,000 obtained and used, with other internal funds, to purchase a Biorad MRC 600 confocal microscope installed late 1991.

Alzheimer's Association Pilot Grant; "Neuronal CDC2-like kinase (NCLK) in the normal and diseased nervous system" G. Shaw P.I. funding 12/1/1993-11/30/1994 $25,000.

Department of Sponsored Research Development Award (University of Florida); "The pleckstrin homology domain". G. Shaw P.I., funding 5/1/1994-4/30/1995 $18,500.

Brain and Spinal Cord Injury Rehabilitation Trust Fund. "Novel Antibodies Recognizing Human Protein Markers" Dr. Shaw P.I. (5% time/effort). Funding from 6/1/1996-5/30/1997, $41,284.

Abbott labs Post-doctoral fellowship to Dr. Deng-Shun Wang, 3/1/1996-2/28/1997 sponsored by G. Shaw, $27,000.

American Cancer Society "The role of the pleckstrin homology domain in cancer". G. Shaw P.I. (10% time/effort) 6/1/1997-5/31/1998 $20,000.

Research and technology investment fund proposal (UF). "Generation of antibodies for commercial purposes". P. I. G. Shaw, started December 1 1998, total funding at $50,000.

American Heart Association (Florida Affiliate) Grant in Aid "The function and regulation of the pleckstrin homology domain of the β-adrenergic receptor kinase." Principle Investigator G. Shaw, (10% time/effort), July 1 1998- June 30 2002, $220,000 total costs.

McKnight Foundation. "Age Related changes in the regulation of MARCKS" P.I. Gerry Shaw, Mike Bubb Co P.I. (10% time/effort) 12/01/02-11/31/03 $75,000 total costs.

DOD/U.S. Army Medical Research and Materiel Command proposal "Common mechanisms of neuronal cell death after exposure to diverse environmental insults: Implications for treatment". Brain Pike P.I., Gerry Shaw Co P.I. (10% Time/effort). 4 years funding from 3/21/2001 to 3/20/2005. Total direct costs $1,523,793.

NIH/NIAAA "Ethanol and Bcl-2 Gene Interactions in Developing CNS". P. I. Marieta Heaton (UF Neuroscience), Co P. I. Gerry Shaw (25% time/effort). 5 years funding from 7/1/01 to 6/30/06 requested. $958,000 total direct costs.

Allan and Maria Myers Travel Fund for research, seminars and teaching in the Howard Florey Institute, Melbourne, Australia $30,000, August-September 2006.

ALS Association "pNF-H as a biomarker of onset of disease in rodent ALS models and in human ALS patients" Gerry Shaw PI (10% Time/effort). 1 August 2006-31 July 2007. $79,000.

ALS Association, "pNF-H as a biomarker of onset and progression of ALS ", essentially a continuation of the above grant for 3 more years, 1 August 2007- 31 July 2010. $80,000 per year.

Department of Veteran Affairs "Locomotor training after spinal cord injury" and "Mechanisms of retroviral injury to the CNS", PI Paul Hoffman, Gerry Shaw Co-PI (12.5% Time/effort) (November 2006 to November 2008, renewed yearly).

Department of Veteran Affairs Merit Review "Serum Biomarkers of Cervical Spinal Cord and Traumatic Brain Injury". P.I. Dena Howland, Co-PI Gerry Shaw (10% time/effort). February 2007 - January 2009. $200,000 total direct costs.

National Institutes of Health "Leber Hereditary Optic Neuropathy:Gene Therapy Trial", R24, P.I. William Hauswirth, Co P.I,. Gerry Shaw (10% time/effort). August 1, 2008 – July 31 2013.

National Institutes of Health "Experimental Optic Neuritis: Gene Therapy", RO1, P.I. John Guy, Gerry Shaw subcontract through EnCor Biotechnology Inc. August 1. 2008 – July 31 2013.

Income to my university research program from university investments, overheads, licensing fees etc.

University of Florida Research Foundation Royalties, under a series of invention disclosures, (now covering more than 60 antibodies, both monoclonal and polyclonal, as well as 2 research and diagnostic kits), P.I. G. Shaw, returns to P.I. laboratory ~$200,000 to date.

Commercial Connections

Numerous antibodies I have produced or helped produce are now commercially available. Monoclonal antibodies to neurofilament L, M, phosphorylated H and phosphate independent H, produced in Göttingen and extensively characterized by myself (refs. 21, 33) are now sold by a variety of companies including Sigma, Amersham and Boehringer-Mannheim. Further monoclonal and polyclonal antibodies to neurofilament triplet proteins, peripherin, α-internexin, GFAP, ubiquitin, glyceraldehyde-3-phosphate dehydrogenase and β-adrenergic receptor kinase 1 produced and characterized in my laboratory in Gainesville (refs 23, 33, 42, 44) are now sold by Abcam, Abnova, Affinity Bioreagents, Chemicon, Fisher, Neomarkers, Novus, Millipore, Neuromics, Novocastra, Santa Cruz, Sigma-Aldrich, Stem Cell Technologies, Thermo-Fisher Scientific, Upstate Biotechnology Inc., Zymed and several other companies. Further antibodies to several molecules involved in signal transduction and cytoskeletal function have also been produced, and many of these have also been made commercially available.

In December 1999 I started up my own small biotechnology company, initially called ABC Biologicals Inc., to exploit some of these immunoreagents and to create new ones. This company was renamed EnCor Biotechnology Inc. and from December 2002 has occupied lab space at the Sid Martin Biotechnology Incubator, Alachua, Florida. This company is commercializing already available antibody reagents and produces novel antibodies and antibody based diagnostic kits. The company achieved profitability in 2004, and profits have increased every year since then. The company moved to a new facility in Gainesville in October 2006. Apart from direct sales of antibodies and ELISA kits, the company also has income from running our increasing panel of biomarker assays on blood and CSF samples from other biotechnology companies.

Patents

In March 2003 I filled a U.S. provisional patent, and a Utility application 60/459,286 "Assessing neuronal damage from blood samples", through the University of Florida. This was followed in March 2004 with the Utility application 10/810,388 with the same title. This was published on the USPTO web site as 2004-0241762 A1 in December 2004. This patent covers the detection of neurofilament subunits as serum and CSF biomarkers of neuronal injury. My collaborator on this work, listed as a co-inventor in the patent, is Dr. Brian Pike, now at the NIH. This application, if approved, will likely form the basis for other more specific applications.

Other Intellectual Property

In 1992 I wrote FINDER, a program which can match the amino acid composition determined experimentally from a PVDF blot of an unidentified protein to proteins in the current protein sequence database which have similar composition (ref. 38). The program also prints out the calculated molecular weight, isoelectric point and name of each candidate. Surprisingly, this program is extremely sensitive and allows rapid and inexpensive identification of unidentified proteins, assuming the unknown protein or a close homologue is found in the protein sequence database, an increasingly likely possibility these days. This program has recently been used to rapidly and inexpensively identify a variety of proteins in my own and other laboratories. Interactive programs based on this approach have recently been made available on the world wide web from two major molecular biology servers, the ExPasy server from the Geneva Hospital and the University of Geneva (http://expasy.hcuge.ch/) and the EMBL server (http://www.embl-heidelberg.de/aaa.html).

I have also written a program called MOTIFER which is very useful for identifying proteins and nucleic acids sharing loosely defined sequence motifs. MOTIFER is designed to accept any defined

range of amino acids at each position, and also tolerating any predefined range in the spacing between amino acids. It has found proteins sharing motifs which were overlooked by the standard FASTA, FASTP and BLAST methods. An example of this is the identification of the only Src-homology 2 (SH2) domain found in yeast, providing an insight into the evolutionary origin of this important signaling module (ref. 43). It was recently shown that this now widely accepted SH2 domain binds specifically to a serine phosphorylated target, which makes some sense as yeast are not known to have tyrosine kinase activities (Yoh et al. Genes Dev. 21:160-174 2007). The same approach identified several examples of previously unrecognized pleckstrin-homology (PH) domains (ref. 39). All of these identifications proved to be correct and led to one of the first theories concerning PH domain function (refs. 39, 46).

I have also described novel methods of analyzing protein and nucleic acid sequences using the surprisingly powerful features built into Microsoft Excel and Microsoft Word. Excel can perform most of the functions one would expect to find in a commercial sequence analysis program, as was originally reported in a Biotechniques paper in 1995 (ref. 50). This approach generated considerable interest and led to an invitation to write a book chapter on the same topic (ref. 57). Recently the original paper was selected for reprinting in a collection of Biotechniques articles along with a short article describing further modifications of this approach (ref. 70). Microsoft Word can also be used to analyze a variety of parameters of protein and nucleic acid sequences by means of Visual Basic macros (ref. 62). A web page has links to all these programs (http://www.mbi.ufl.edu/~shaw/comp.htm).

Finally I learned Javascript and Java, two powerful programming languages which are particularly applicable to web page design. Javascript can perform quite complex mathematical calculations and Javascript code can be embedded in web pages, allowing the host computer to run these programs. I have written various useful programs which are now freely available on my company internet site at http://www.encorbio.com/protocols.htm. Many of these protocols and calculators are linked to by other internet sites such as Protocols-online, Biocompare.com and Wikipedia. Javascript has a very limited ability to display complex graphics, so I am currently producing more pages using Java, which can also be embedded in web pages, but has much greater ability to display complex graphics.

Referees- (others on request)

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